

Urolithiasis in the maned wolf (*Chrysocyon brachyurus*): Assessment of four clinical cases in captivity

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Abstract

Four cases of urolithiasis in captive maned wolves (*Chrysocyon brachyurus*) were studied. All four animals were adult males. The clinical cases occurred in the period 1989–2004. The disease was manifested after 2 months to 10 years of captivity. The main clinical symptoms found were abdominal distension, recurrent urinary tract infections, pain upon abdominal palpation, difficulty urinating, polyuria, hematuria, anorexia, dehydration, and urinary tenesmus evolving to anuria due to obstruction of the urethra by calculi. Radiological examination detected bladder distension and the presence of many radio-opaque uroliths in the bladder lumen. Diffraction analysis of uroliths showed they were made up of calcium acid pyrophosphate and basic hydrous manganese phosphate (n=1) and hydrous magnesium ammonium phosphate with traces of Potassium Calcium Phosphate (n=1). Electron microscopy showed that uroliths consisted of crystals with mineral phosphorus, potassium and magnesium (n=2) predominating in their composition. This is the first time any scientific research has demonstrated the occurrence of urolithiasis associated with uroliths of mineral composition based on phosphate, potassium and magnesium in captive maned wolves. The only cases of urolithiasis in maned wolves documented hitherto were cystine-related. Therapeutic measures for urolithiasis involve appropriate nutritional management, use of medication and, in some cases, specific surgical procedures. Diagnosis and treatment of the diseases to which wild animals are subject when in captivity are extremely important for maintenance and reproduction of these species with a view to their conservation in the environment.

Key words:

Maned wolf.
Urolithiasis.
Mineral composition
of uroliths.
Clinical symptoms of
urolithiasis.

Introduction

The maned wolf, *Chrysocyon brachyurus* (Illiger 1811), is a carnivorous mammalian of the Canidae family which inhabits areas from the Northeast of Brazil to northern Argentina, Paraguay, eastern Bolivia and western Peru.¹ In Brazil it is officially classified as an endangered species under Ministry of the Environment Normative Instruction 03/2003. It is a solitary animal with crepuscular to nocturnal activity patterns. In the wild it feeds mostly on small vertebrates, fruits and

berries.² Urolithiasis is not a specific disease but secondary to underlying disorders. It is characterized by the formation of stones or calculi (uroliths) in the kidneys, ureters and/or bladder. Uroliths are formed when there is an increase in the conditions that favors their development, such as urinary tract infections, unbalanced diet and genetic predisposition.³ The purpose of this study was to assess the clinical, radiological and histopathological profiles of urolithiasis in maned wolves kept in captivity; to analyze urolith composition qualitatively and

quantitatively; and to propose strategies for early diagnosis of the disease as well as suitable treatment for affected animals.

Material and Method

Four cases of urolithiasis in maned wolves were studied. All four cases occurred in the period 1989-2004 at the Captive Breeding Compound, a unit of Companhia Brasileira de Metalurgia Mineração - CBMM's Environmental Development Center near Araxá, Minas Gerais, Brazil. Data were analyzed and compiled from clinical records and autopsy reports on diseased animals. Uroliths found during clinical or postmortem examination were analyzed at CBMM's Research Center, by X-ray diffraction (XRD), using a Rigaku Geigerflex, and scanning electron microscopy (SEM), using a Cambridge Instruments S200 with EDS. For XRD, each urolith sample was carefully ground into a powder and loaded into a sample port. For the SEM analysis, uroliths were first dried at 60°C and then metallized with carbon.

Results

The analyses showed that clinical cases occurred in four adult males, with three resulting in death. Half the animals originated from the wild ($n=2$) and half were born in captivity ($n=2$). The disease was manifested after 2 months to 10 years of captivity. In one case ($n=1$), the animal presented with acute incontinence, which eventually resulted in death due to bladder wall perforation by a single pointed calculus with 4 cm in length by 1.5 cm in width (Figure 1). In two cases ($n=2$), recourse to surgery revealed similar clinical features that were evidently chronic, with urinary tract infection, concurrent formation of several uroliths of various sizes, and irreversible damage making euthanasia unavoidable (Figure 2). One of the animals ($n=1$) has survived and is now undergoing treatment at the Breeding Compound. The main clinical symptoms found were abdominal distension ($n=3$),

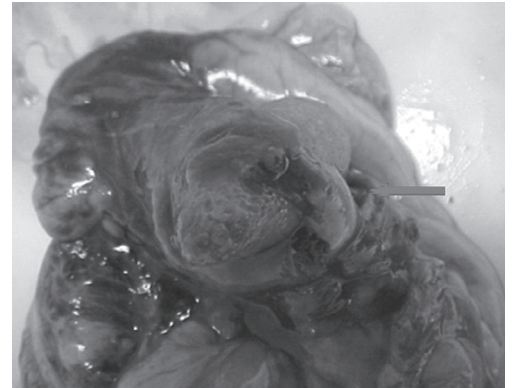


Figura 1 - Bladder wall perforation by a single pointed calculus with 4 cm in length by 1,5 cm in width found in a maned wolf (*Chrysocyon brachyurus*), during postmortem examination at the Captive Breeding Compound, Araxá - MG - Brazil. The stone was composed by $\text{MgNH}_4\text{PO}_4 \cdot 6\text{H}_2\text{O}$ - hydrous magnesium ammonium phosphate with traces (1%) of KCaPO_4 - potassium calcium phosphate

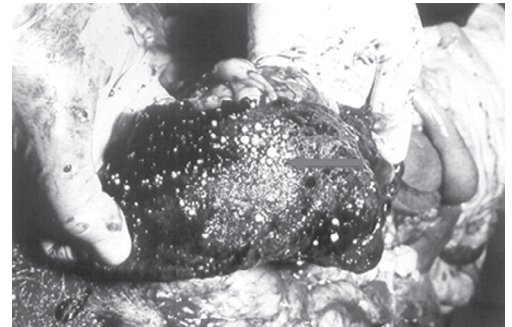


Figura 2 - Several stones encrusted in the bladder wall found in a maned wolf (*Chrysocyon brachyurus*) during postmortem examination at the Captive Breeding Compound, Araxá - MG - Brazil. The stones were composed by $\text{CaH}_2\text{P}_2\text{O}_7$ - calcium acid pyrophosphate and $\text{MnFe}_2(\text{PO}_4)_2(\text{OH}) \cdot 2\text{H}_2\text{O}$ - basic hydrous manganese phosphate

recurrent urinary tract infections ($n=3$), pain upon abdominal palpation ($n=3$), difficulty urinating ($n=4$), polacuria ($n=3$), hematuria ($n=2$), anorexia ($n=2$), dehydration ($n=2$), and urinary tenesmus coming to anuria due to urethra obstruction by calculi ($n=2$). Radiological examination of three animals detected bladder distension and the presence of many radio-opaque uroliths in the bladder lumen of two animals ($n=2$). Uroliths found in three animals displayed a rounded shape, varying sizes and a yellowish color. Diffraction analysis of the uroliths showed they were made up of $\text{CaH}_2\text{P}_2\text{O}_7$ - calcium acid pyrophosphate and

$\text{MnFe}_2(\text{PO}_4)_2(\text{OH}) \cdot 2\text{H}_2\text{O}$ - basic hydrous manganese phosphate (n=1) and $\text{MgNH}_4\text{PO}_4 \cdot 6\text{H}_2\text{O}$ - hydrous magnesium ammonium phosphate with traces (1%) of KCaPO_4 - potassium calcium phosphate (n=1). Electron microscopy showed that uroliths consisted of crystals with mineral phosphorus, potassium and magnesium (n=2) predominating in their composition (Figures 3 and 4). On postmortem examination, the three deceased animals were found to have several calculi encrusted in the bladder wall, in addition to a swollen and distended bladder. In one case, a histopathological examination of the kidneys



Figura 3 - Scanning electron micrograph (100 X) of the urolith found in one maned wolf (*Chrysocyon brachyurus*) during clinical examination at the Captive Breeding Compound, Araxá - MG - Brazil. Panoramic view showing the internal part of the urolith, which was composed by crystals. Secondary Electron (SE), scale bar = 100 µm, accelerating voltage 25,0 kV

and bladder showed chronic pyelonephritis and a nonspecific chronic inflammatory process with pronounced activity in both the mucosa and the wall of the urinary vesicle.

Discussion

In the present study the clinical cases were observed only in males, suggesting a sex-related pattern, possibly due to the anatomy of the maned wolf's penile urethra, which is long and narrow and also because

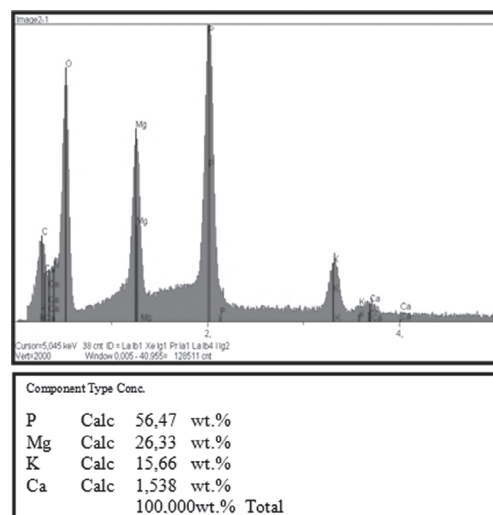


Figura 4 - Graphic of the chemical composition of the uroliths, by scanning electron microscopy (SEM), that micrograph was showed in figure 3. The SEM showed that uroliths were made by phosphorus, magnesium, potassium and calcium

the maned wolf has a penile bone. According to the literature, an important health problem for maned wolves is the formation of cystine calculi leading to the formation of stones in the kidney, bladder and urethra. A study of maned wolves in captivity and in the wild concluded that 80% of the animals concerned were capable of excreting cystine and other dibasic amino acids in their urine. It is believed that this unusual pattern may be due to a metabolic disturbance of genetic origin.⁴ Urinary calculi composed of cystine were found in four maned wolves and proved to be fatal in three of them.⁵ Mussart and Coppo⁶ reported finding of six cystine calculi incrustated in the renal pelvis of and adult *Chrysocyon brachyurus* whose death was preceded by respiratory problems. Basically three theories have been proposed to explain the physiopathology of the urolithiasis: Precipitation-crystallization theory, Nucleation hypohesis and Crystallization inhibition theory. Whichever the theory involved in the physiopathology, an important condition that the urine must fulfill is the unstable oversaturation.⁷ Because urine is commonly supersaturated, observation of crystals in the urine does not mean the patient is at risk for urolithiasis. Supersaturation of urine depends

on the amount of the ion ingested and excreted in the volume of urine produced.⁸ The urine must be oversaturated with minerals and these have to rely on suitable physical and chemical conditions that should facilitate the precipitation.⁷ Most stones have one major crystal component, which can be formed for several possible reasons, such as unbalanced diet, decreased water intake, modified urine pH, relative lack of inhibitors of crystallization. For these reasons the solubility product of a particular crystal may be exceeded, crystals may form and these crystals may aggregate and grow. The most common stones in dogs and cats are struvite, calcium oxalate and calcium phosphate, urate, cystine and silicate.⁸

In the present study, a calculi composed by $\text{MgNH}_4\text{PO}_4 \cdot 6\text{H}_2\text{O}$ with traces of KCaPO_4 was formed in one maned wolf and identified as the composition of struvite stones.⁸ In dogs, struvite urolithiasis is predominately a disease associated with infection. Bacteria promote increased ammonium concentration creating a highly alkaline pH. Alkaline pH with urine saturated with ammonium, phosphate and magnesium ions results in nucleation of struvite crystals.⁹ In dogs which struvite stones are formed in spite of the absence of urinary tract infection, predisposing factors include a family history of struvite stones, a diet based on vegetable proteins and distal renal tubular acidosis.⁸ In cats, the risk of struvite uroliths increases due to dietary influences on urinary pH and stone-forming constituents (i.e. magnesium, phosphorus and ammonium) and urinary tract infection are not the driving force behind the formation of alkaline urine.⁹ The struvite stones are spherical, ellipsoidal or tetrahedral in shape and may be present in singly or in large numbers of varying sizes. The bladder is the most common site of struvite stone formation, although it can occur at any site in the urinary tract.⁸

Another calculus composed by calcium and phosphate and manganese phosphates was found in one maned wolf. Calcium oxalate or calcium phosphate calculi are usually white in colour and very hard.

They often have sharp, jagged edges, may be single or in multiple number and are found most often in the bladder and urethra. Urinary tract infection can be a predisposing factor to calcium oxalate urolithiasis. Altered calcium metabolism also may play a role in development of calcium oxalate urolithiasis.⁸

In the present study it was observed that as well as occurring in association with recurrent urinary infections, urolithiasis may also have been caused by unsuitable diet, especially in light of the fact that the nutritional requirements for captive maned wolves are unknown.¹⁰

The uroliths damage the urinary tract mucous, which generates inflammation (hematuria, pollakiuria, dysuria, stranguria) and it predisposes to the bacterial colonization and infection of urinary tract. The clinical signs depend on the anatomical location, antiquity and physical characteristics of the uroliths precedents from the urethral acute partial or complete obstruction. The patients can be asymptomatic when the uroliths are in the unilateral renal pelvis or at the ureters, or to present hematuria and abdominal pain causing often hydronephrosis.⁷ In the present study, the majority of the symptoms were observed only in the final clinical stage of the disease.

The assessment of uroliths presence is done by simple radiographic studies or with contrast.⁷ Appropriate survey or contrast radiography or ultrasonography should always be performed to localize the site (s) of obstruction, as well the number, size and surface characteristics of the uroliths. Although survey abdominal radiography will not detect all uroliths, the majority of the common uroliths (struvite, calcium oxalate, cystine) are radiodense to some degree, with the exception of some urate uroliths that are radiolucent.¹¹ According to Gallatti and Iwasaki¹², the positive contrast-cystography is more efficient in detecting mucosal surface irregularity of the bladder, bladder displacement and intraluminal structures, except calculi, and the ultrasonography is more efficient in detecting uroliths and bladder wall thickening. The urinalysis is a

useful tool for the diagnosis of crystalluria, to determine the type of mineral that saturates the urine and represents part of the composition of the urolith. The determination of the pH through the urinalysis is also important for differential diagnosis of the uroliths, as well as some other general finds as hematuria or pyuria.⁷ Calculi composed by struvite, calcium carbonate, calcium phosphate and urate are more soluble in acidic urine (pH < ~ 6.8); uric acid and cystine are more soluble in alkaline urine (pH > ~ 7.0); and calcium oxalate are minimal affected by urine pH.⁸ The macroscopic morphology is too diverse as for to allow a diagnosis based on the physical aspects of the stones. It is necessary to carry out a correct analysis of each calculi to determine the types of mineral of which they are constituted to establish the treatment for the animal.⁷

The treatment of urolithiasis is based on the relief of urinary tract obstruction and reestablishment of urine flow.⁸ Urethral obstruction is a life-threatening situation and can be fatal due to renal dysfunction and retention of metabolic wastes, especially potassium and metabolic acids.¹¹ Strategies for treating struvite stones in dogs involve treating the urinary tract infection in conjunction with physical or dietary removal of existing stones.⁹ The use of urinary acidifiers to maintain urine pH in the range of 6.0-6.5 has been suggested in dogs because struvite and hydroxyapatite are most soluble in acidic urine.⁸ In dogs, the dietary changes for struvite prevention includes promotion of an acid urine (pH 6.5) together with phosphorus and magnesium restriction.⁹ Surgery is required to remove calcium oxalate stones in dogs and cats. Postoperatively, a diet low in calcium and oxalate

should be fed. Dietary phosphorus should not be restricted because reduced phosphorus could result in increased activation of vitamin D3 to calcitriol by 1- α -hydroxylase in the kidney and cause increased intestinal absorption of calcium. Also, urinary pyrophosphate may function as an inhibitor of calcium oxalate formation. Dietary magnesium should not be restricted either, because it may serve as an inhibitor of calcium oxalate formation.⁸

Conclusion and Recommendations

This is the first time any scientific research has demonstrated the occurrence of urolithiasis associated with uroliths of mineral composition based on phosphate, potassium and magnesium in captive maned wolves. The only cases of urolithiasis in maned wolves documented hitherto were cystine-related. Moreover, analysis of the results showed a high rate of mortality correlated with urolithiasis in this species, pointing to a need to develop a protocol involving periodic clinical assessment supplemented by biochemical tests and diagnostic imaging to detect early onset of the disease for providing suitable treatment in time and prevent complications, which if not treated in a timely and adequate manner may result in death. Therapeutic measures for urolithiasis involve appropriate nutritional management, use of medication and, in some cases, specific surgical procedures. Diagnosis and treatment of the diseases to which wild animals are subject when in captivity are extremely important for maintenance and reproduction of these species with a view to their conservation in the environment.

Urolitíase em lobo guará (*Chrysocyon brachyurus*): Avaliação de quatro casos clínicos em cativeiro

Resumo

Foram estudados quatro casos de urolitíase em lobos guarás (*Chrysocyon brachyurus*), que ocorreram no período de 1989 a 2004, de

Palavras-chave:

Lobo Guará. Urolitíase. Composição mineral de urólitos. Sintomas clínicos da urolitíase.

animais mantidos em cativeiro. Os casos clínicos ocorreram em quatro machos adultos. O tempo de cativeiro para a manifestação da doença nos animais variou de dois meses a 10 anos. Os principais sintomas clínicos apresentados foram distensão abdominal, infecções recorrentes do trato urinário, dor à palpação abdominal, dificuldade em urinar, polaciúria, hematúria, anorexia, desidratação e tenesmo urinário com evolução para anúria em decorrência da obstrução da uretra por cálculos. Ao exame radiológico detectou-se distensão da bexiga e a presença de inúmero urólitos radiopacos no lúmen do órgão. As análises difratométricas dos urólitos revelaram que eles eram compostos de Pirofostato Ácido de Cálcio e Fosfato Básico de Manganês Hidratado (n=1)) e Fosfato de Amônio Magnésio Hidratado com traços de Fosfato de Potássio e Cálcio (n=1). A microscopia eletrônica revelou que os urólitos eram formados por cristais, com predominância dos minerais de fósforo, potássio e magnésio (n=2) em sua composição. O presente estudo demonstrou, de forma inédita, a ocorrência de urolitíase associada a urólitos de composição mineral a base de fosfato, potássio e magnésio, em lobos guarás cativos. Até então, só foram documentados casos de urolitíase em lobos guarás relacionados à cistina. As medidas terapêuticas da urolitíase envolvem manejo nutricional adequado, utilização de medicamentos e, em alguns casos, cirurgias específicas. O diagnóstico e tratamento das doenças que acometem animais silvestres em cativeiro são de extrema importância para a manutenção e reprodução dessas espécies, visando à sua conservação no meio ambiente.

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